REMARKS/ARGUMENTS

With this amendment, claims 1, 6, 14-16, 19, and 28-31 are pending. For convenience, the Examiner's rejections are addressed in the order presented in the January 11, 2006, Office Action. Applicants thank Examiners Chan and Haddad for taking time to participate in a telephonic interview on April 11, 2006, with Applicant's representatives Beth Kelly, Annette Parent and James Diehl. The rejections under 35 U.S.C. §112, first paragraph, alleged lack of written description and enablement were discussed. No agreement was reached.

I. Status of the claims

Claim 1 is amended to recite that the ILKAP protein has an anti-angiogenic phenotype. Support for this amendment is found throughout the specification, for example at page 3, lines 29-34; page 29, lines 3-10, and Example 1, page 44-45. This amendment adds no new matter.

I. Rejections under 35 U.S.C. §112, first paragraph, enablement

Claims 1, 6, 14-15, 19, and 28-30 are rejected under 35 U.S.C. §112, first paragraph because allegedly, the specification does not provide enablement for one of skill to make and use an invention commensurate in scope with those claims, *i.e.*, methods to use an ILKAP protein with 90% identity to SEQ ID NO:2. The Office Action also alleges that undue experimentation is required to practice the claimed invention. To the extent the rejection applies to the claims as amended, Applicants respectfully traverse the rejection.

In order to establish a prima facie case of lack of enablement, the Examiner has the burden to establish a reasonable basis to question the enablement provided for the claimed invention. *In re Wright*, 27 USPQ2d 1510, 1513 (Fed. Cir. 1993). The Examiner has not provided any reason why those of skill would not be able to practice the claimed methods based on the disclosure of the specification and on information that was publicly available at the time of filing.

The Office Action in large part alleges that undue experimentation is required to practice the invention. As set forth in the Manual of Patent Examining Procedure (MPEP) §2164.01, "the test of enablement is not whether any experimentation is necessary, but whether… it is undue." Further, the "fact that experimentation may be complex does not necessarily make it undue, if the art typically engages in such experimentation" (citations omitted). Finally, claims reading on inoperative embodiments are enabled if the skilled artisan understands how to avoid inoperative embodiments. *See, e.g., In re Cook and Merigold*, 169 USPQ 299, 301 (C.C.P.A. 1971). Moreover, "[a] patent need not teach, and preferably omits, what is well known in the art." MPEP 2164.01 *citing In re Buchner*, 18 USPQ2d 1331, 1332 (Fed. Cir. 1991); *Hybritech, Inc. v. Monoclonal Antibodies, Inc.*, 231 USPQ 81, 94 (Fed. Cir. 1986), *cert. denied*, 480 U.S. 947 (1987); *Lindemann Maschinenfabrik GMBH v. American Hoist & Derrick Co.*, 221 USPQ 481, 489 (Fed. Cir. 1984).

Ex parte Sun et al. and Ex parte Bandman et al.

In the previous response, Applicants cited *Ex parte Sun et al.*, an opinion by the Board of Patent Appeals and Interferences, in support of enablement of pending claims that recite 90% or 95% identity to a reference sequence. The Office Action asserts that Applicants mischaracterized the Board's decision in *Sun*. Applicants respectfully remind the Examiner that in *Sun*, the Board found claims that recited 80% identity to a reference sequence (a WEE1 protein) were enabled. In the present Office Action, the rejections and reasoning applied the WEE1 claims during prosecution are cited against the pending claims, *i.e.*, that alteration of a critical region of the WEE1 protein could affect protein function and therefore, that one of skill would not be able to predict the structure and function of sequence variations of the protein. Applicants respectfully assert that the prosecution reasoning in *Sun* was actually rejected by the Board, who ruled that the Examiner **did not** demonstrate that the teachings of the application at issue were insufficient for enablement of the claims to 80% identity to a reference sequence. In the present Office Action, Examiner Haddad has not demonstrated that the evidence presented in *Sun* is somehow more enabling than the evidence relied on by the Applicants in the present case.

Therefore, the citation of *Sun* supports the present Applicants assertion of enablement for claims that recite 90% identity to a reference sequence.

Applicants also bring to the Examiner's attention another recent decision by the Board of Patent Appeals and Interferences: *Ex parte Bandman*, Appeal No. 2004-2319. Using reasoning similar to that of *Sun*, in *Bandman*, the board found that claims directed to sequences with 95% identity to a reference sequence were enabled because the supporting specifications provided a single reference sequence and an assay for activity of the encoded protein.

The Office Action appears to assert that amino acid by amino acid investigation of structure function relationships is required for enablement of claims that recite, *e.g.*, 90% identity to a reference sequence. Both *Sun* and *Bandman* reject that analysis and put forth an enablement standard of identification of functional areas in the amino acid sequence of a protein combined with assays to measure activity of the protein.

Evidence in support of Enablement

According to the Office Action, the declaration of Dr. Sacha Holland, submitted with the previous response, presents only the opinions of Dr. Holland and no factual evidence of enablement of the recited ILKAP polypeptides. Applicants dispute this finding.

Dr Holland's declaration provided explanation of references and other evidence of the knowledge of those of skill at the time of filing. The declaration of Dr. Holland provides a summary of the knowledge of the ILKAP proteins and related proteins at the time of filing, *e.g.*, assays of phosphatase activity and regions of conservation between related phosphatases. Moreover, such statements are supported by published references that were also submitted with the previous response. *See, e.g.*, Leung-Hagesteijn *et al*., *EMBO J.*, 20:2160-2170 (2001) and Das *et al*., *EMBO J.* 15:6798-6809 (1996). Dr. Holland also pointed out support in the specification for assays to measure the anti-angiogenic function of the ILKAP protein. Finally, Dr. Holland corrected misinterpretations of references cited in the previous office action. The Office Action did not address the evidence, but rather asserted that the declaration was unsupported and uncorroborated and thus deficient. Applicants assert again, based on Dr.

Holland's declaration and the supporting references and information in the specification, that those of skill in the art would know how to identify ILKAP proteins with 90 % identity to the reference sequence that have the recited anti-angiogenic activity.

Applicants also dispute the Office Action's characterization of the knowledge of the function of the ILKAP protein at the time of filing and of two of the cited references: Atwood and Skolnick. First, the Office Action at page 6 asserts that ILKAP was known only as a protein without a function and that the specification allegedly discloses that structural similarity among phosphatase is not predictive of functional similarity. However, as described below, the specification discloses both the identification and functional characterization of the ILKAP protein that occurred before the filing date. The specification discloses that nucleic acid encoding the ILKAP protein was first disclosed in GenBank and did not include a function for the protein. But the specification also discloses published empirical evidence of a function for the ILKAP protein, *i.e.*, phosphatase activity and regulation of the ILK protein, in Leung-Hagesteijn *et al*. According to the MPEP, the Examiner must weigh all evidence to establish a reasonable basis to question enablement. MPEP 2164.05. In limiting the analysis to the first GenBank disclosure, the Examiner has failed to consider all the evidence. Therefore, the Office Action's characterization of the knowledge of the ILKAP protein at the time of filing does not meet the standard to establish a reasonable basis to question enablement.

Based on the incorrect characterization, the Office Action at page 6 asserts that the ILKAP protein and variants allegedly are a polypeptides whose function cannot be predicted and must be determined empirically. Applicants respectfully remind the Examiner that, in addition to the published phosphatase activity of the ILKAP protein, the specification discloses assays for the angiogenic activity of the ILKAP protein. Thus, those of skill can carry out at least two assays for ILKAP function: angiogenic assays and phosphatase assays. There is no requirement in the cited references to empirically determine additional functions for the ILKAP protein or its variants. Moreover, ILKAP variants that do not have the now-claimed anti-angiogenic phenotype will not be covered by the claims.

Undue experimentation

According to the Office Action undue experimentation is required from those of skill to practice the claimed methods using proteins with 90% or 95% identity to SEQ ID NO:2. Applicants respectfully traverse. The claimed methods can be practiced by those of skill with, at most, routine experimentation.

The Office Action appears most concerned by claims directed to ILKAP polypeptides that have 90% or 95% identity to a reference sequence, asserting that as many as 39 amino acid substitutions could be included and that the expectation that any "given artificially synthesized polypeptide that is 90% identical to SEQ ID NO:2 would be functional is astronomically low." Applicants respectfully remind the Examiner that molecular biologists routinely screen large numbers of candidate molecules for a nucleic acid or protein associated with a phenotype of interest. Assays to identify ILKAP polypeptides that have an anti-angiogenic phenotype and that are 90% identical to SEQ ID NO:2 are disclosed in the specification and can be routinely performed by those of skill, even in "astronomically" large numbers.

The Federal Circuit Court of Appeals has issued a number of decisions that put forth the standard for undue experimentation in the field of molecular biology. Complex experiments are not necessarily undue, so long as they are typical of the art. *See, e.g., In re Certain Limited-Charge Cell Culture Microcarriers*, 221 USPQ 1165, 1174 (US ITC, 1983). The quantity of experimentation is not dispositive and a "considerable amount of experimentation is permissible, if it is merely routine. . . " *See, e.g., In re Wands*, 8 USPQ2d 1400, 1404 (Fed. Cir. 1988). Most recently, the Federal Circuit reviewed a Board of Patent Appeals and Interferences decision on enablement of claims to recombinant pox viruses and agreed that "the mere fact the that experimentation may have been difficult and time consuming does not mandate a conclusion that such experimentation would have been considered to be 'undue' in this art." *See, e.g., Falkner v. Inglis*, 79 USPQ2d 1001, 1006 (Fed. Cir. 2006).

The ILKAP polypeptide was identified as a target for modulation of angiogenesis using a flow cytometry screen to measure differences in levels of the αvβ3 protein, an

angiogenesis marker, after transduction of an appropriate library into endothelial cells. Fluorescently labeled anti- αvβ3 antibodies were used to assay protein levels and decreased αvβ3 protein levels indicate inhibition of angiogenesis. Hundreds of thousands of transduced cells were screened in order to identify the ILKAP functional hit. It is well known that, using flow cytometry, those of skill can assay 5000 cells per second. *See, e.g.*, Exhibit A, Alberts *et al.*, *Molecular Biology of the Cell*, 157-158 (1994). Thus, the disclosed flow cytometry assays can easily be used to screen large number of candidates for proteins with 90% identity to SEQ ID NO:2 and the recited anti-angiogenic function. Flow cytometry assays are routinely performed by molecular biologists and daily screens of several thousands to millions of potential molecules are also routinely performed by molecular biologists. Thus, any experimentation required to identify an allegedly rare functional ILKAP polypeptide with 90% identity to SEQ ID NO:2 would not be undue.

In view of the above arguments and amendments, withdrawal of the rejection for alleged lack of enablement is respectfully requested.

II. Rejections under 35 U.S.C. §112, first paragraph, written description

Claims 1, 6, 14-15, 19, and 28-30 are rejected under 35 U.S.C. §112, first paragraph for allegedly failing to comply with the written description requirement. According to the Office Action, the specification does not provide description of polypeptides with at least 90-95% identity to SEQ ID NO:2. The Office Action alleges that those of skill would not recognize that the inventors had possession of the claimed invention at the time of filing. The Office Action addresses the analysis to polypeptides that have 90% identity to SEQ ID NO:2. Applicants respectfully remind the Examiner that certain dependent claims are directed to polypeptides that have 95% identity to SEQ ID NO:2 and that those dependent claims should also be considered.

Applicants respectfully traverse the rejection. As currently applied, the specification does comply with US patent law for description of a nucleic acid or amino acid

sequence. The Federal Circuit court of Appeals addressed the description adequate to show one of skill that the inventors were in possession of a claimed genus at the time of filing. *See, e.g., Enzo Biochem, Inc. v. Gen-Probe, Inc.*, 63 USPQ2d 1609 (Fed. Cir. 2002). An applicant may also show that an invention is complete by

> . . . disclosure of sufficiently detailed, relevant identifying characteristics which provide evidence that applicant was in possession of the claimed invention . . . *i.e.*, complete or partial structure, other physical and/or chemical properties, functional characteristics when coupled with a known or disclosed correlation between function and structure, or some combination of such characteristics. *Id*. at 1613.

Furthermore, "description of a representative number of species does not require the description to be of such specificity that it would provide individual support for each species that the genus embraces." *See, e.g.*, 66 Fed. Reg. 1099, 1106 (2001).

Functional assays to identify proteins with anti-angiogenic phenotypes (*i.e.*, ILKAP polypeptides) as claimed were known to those of skill and are disclosed in the specification. For example, the specification describes methods of determining an effect on angiogenesis through disclosure of multiple angiogenesis assays. Assays for angiogenesis include assays for expression of cell surface markers, such as $\alpha v\beta 3$ (page 3, lines 29-34; page 5, lines 22-34; page 28, lines 8-11; and exemplified in Example 1 at pages 44-45); haptotaxis assays (page 3, lines 29-34; page 5, lines 22-34; and exemplified in Example 1 at page 45); a chick CAM assay (page 29, lines 14-20); a mouse corneal assay (page 29, lines 21-24); and assays for neovascularization of tumors (page 29, lines 25-29). The specification also discloses the ILKAP protein sequence as SEQ ID NO:2, as well as alignment programs and methods to modify nucleic acids that encode the ILKAP protein.

In *Falkner v. Inglis*, the Federal Circuit ruled that, for claims to nucleic acid sequences and by analogy to amino acid sequences, absence of examples does not render written description inadequate and that actual reduction to practice is not required. *See, e.g.*, *Falkner* at page 1008. The court also ruled that publicly available references that describe essential regions

of a pox virus could be used to allow those of skill to choose an essential vaccinia gene and then to make a claimed virus. *See, e.g.*, *Falkner* at page 1008. In the present case Applicants have identified functionally important regions of the ILKAP protein to assist those of skill in designing polypeptides with 90% identity to SEQ ID NO:2. Applicants again bring to the Examiner's attention the *Sun* and *Bandman* decisions by the Board of Patent Appeals and Interferences. In both cases, the board found that claims directed to sequences with 80% or 95% identity to a reference sequence were enabled because the supporting specifications provided a single reference sequence and an assay for activity of the encoded protein.

Applicants also direct the Examiner's attention to Example 14 of the Synopsis of Application of Written Description Guidelines which analyzes a claim directed to a protein having an amino acid sequence at least 95% identical to SEQ ID NO:3 and that has a specific activity. In these Guidelines, the Patent Office concluded that the claim was adequately described within the meaning of 35 U.S.C. §112, first paragraph. The ILKAP protein does have an anti-angiogenic phenotype as demonstrated at Example 1, pages 44-45. Therefore, on the basis of Written Description Guidelines issued by the USPTO, the present claims directed to sequences that are 95% identical to SEQ ID NO:2, meet the written description requirement.

In view of the above arguments and amendments, withdrawal of the rejection for alleged lack of written description is respectfully requested.

CONCLUSION

In view of the foregoing, Applicants believe all claims now pending in this Application are in condition for allowance. The issuance of a formal Notice of Allowance at an early date is respectfully requested.

If the Examiner believes a telephone conference would expedite prosecution of this application, please telephone the undersigned at 415-576-0200.

Respectfully submitted,

/Beth L. Kelly/

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MOLECULAR BIOLOGY OF
THE CELL
THIRD EDITION

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James D. Watson

EXHIBIT A

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Front cover: The photograph shows a rat nerve cell in culture. It is labeled (*yellow*) with a fluorescent antibody that stains its cell body and dendritic processes. Nerve terminals (*green*) from other neurons (not visible), which have made synapses on the cell, are labeled with a different antibody. (Courtesy of Olaf Mundigl and Pietro de Camilli.)

Dedication page: Gavin Borden, late president of Garland Publishing, weathered in during his mid-1980s climb near Mount McKinley with MBoC author Bruce Alberts and famous mountaineer guide Mugs Stump (1940–1992).

Back cover: The authors, in alphabetical order, crossing Abbey Road in London on their way to lunch. Much of this third edition was written in a house just around the corner. (Photograph by Richard Olivier.)

Cells Can Be Isolated from a Tissue and Separated into Different Types [15]

The first step in isolating cells of a uniform type from a tissue that contains a mixture of cell types is to disrupt the extracellular matrix and intercellular junctions that hold the cells together. The best yields of viable dissociated cells are usually obtained from fetal or neonatal tissues, typically by treating them with proteolytic enzymes (such as trypsin and collagenase) and with agents (such as ethylenediaminetetraacetic acid, or EDTA) that bind, or *chelate*, the Ca^{2+} on which cell-cell adhesion depends. The tissue can then be dissociated into single viable cells by gentle agitation.

Several approaches are used to separate the different cell types from a mixed cell suspension. One involves exploiting differences in physical properties. Large cells can be separated from small cells and dense cells from light cells by centrifugation, for example. These techniques will be described in connection with the separation of organelles and macromolecules, for which they were originally developed. Another approach is based on the tendency of some cell types to adhere strongly to glass or plastic, which allows them to be separated from cells that adhere less strongly.

An important refinement of this last technique depends on the specific binding properties of antibodies. Antibodies that bind specifically to the surface of only one cell type in a tissue can be coupled to various matrices—such as collagen, polysaccharide beads, or plastic—to form an *affinity surface* to which only cells recognized by the antibodies will adhere. The bound cells are then recovered by gentle shaking, by treatment with trypsin to digest the proteins that mediate the adhesion, or, in the case of a digestible matrix (such as collagen), by degrading the matrix itself with enzymes (such as collagenase).

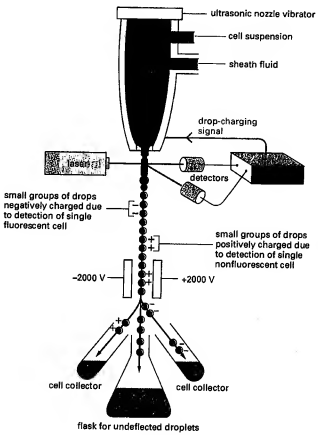

Figure 4–31 A fluorescence-activated cell sorter. When a cell passes through the laser beam, it is monitored for fluorescence. Droplets containing single cells are given a negative or positive charge, depending on whether the cell is fluorescent or not. The droplets are then deflected by an electric field into collection tubes according to their charge. Note that the cell concentration must be adjusted so that most droplets contain no cells and flow to a waste container together with any cell clumps. The same apparatus can also be used to separate fluorescently labeled chromosomes from one another, providing valuable starting material for the isolation and mapping of genes.

The most sophisticated cell-separation technique involves labelling specific cells with antibodies coupled to a fluorescent dye and then separating the labeled cells from the unlabeled ones in an electronic **fluorescence-activated cell sorter.** Here, individual cells travelling in single file in a fine stream are passed through a laser beam and the fluorescence of each cell is measured. Slightly farther downstream, tiny droplets, most containing either one cell or no cells, are formed by a vibrating nozzle. The droplets containing a single cell are automatically given a positive or a negative charge at the moment of formation, depending on whether the cell they contain is fluorescent; they are then deflected by a strong electric field into an appropriate container. Occasional clumps of cells, detected by their increased light scattering, are left uncharged and are discarded into a waste container. Such machines can select 1 cell in 1000 and sort about 5000 cells each second (Figure 4–31).

When a uniform population of cells has been obtained by any of these methods, it can be used directly for biochemical analysis. Alternatively, it provides a suitable starting material for cell culture, allowing the complex behavior of cells to be studied under the strictly defined conditions of a culture dish.

Cells Can Be Grown in a Culture Dish [16]

Given appropriate conditions, most kinds of plant and animal cells will live, multiply, and even express differentiated properties in a tissue-culture dish. The cells can be watched under the microscope or analyzed biochemically, and the effects of adding or removing specific molecules, such as hormones or growth factors, can be explored. In addition, in a mixed culture the interactions between one cell type and another can be studied. Experiments on cultured cells are sometimes said to be carried out *in vitro* (literally, "in glass") to contrast them with experiments on intact organisms, which are said to be carried out *in vivo* (literally, "in the living organism"). The terms can be confusing because they are often used in a different sense by biochemists, for whom *in vitro* refers to biochemical reactions occurring outside living cells, while *in vivo* refers to any reaction taking place inside a living cell.

Tissue culture began in 1907 with an experiment designed to settle a controversy in neurobiology. The hypothesis under examination was known as the *neuronal doctrine,* which states that each nerve fiber is the outgrowth of a single nerve cell and not the product of the fusion of many cells. To test this contention, small pieces of spinal cord were placed on clotted tissue fluid in a warm, moist chamber and observed at regular intervals under the microscope. After a day or so, individual nerve cells could be seen extending long, thin processes into the clot. Thus the neuronal doctrine was validated, and the foundations for the cell-culture revolution were laid.

The original experiments in 1907 involved the culture of small tissue fragments, or **explants.** Today, cultures are more commonly made from suspensions of cells dissociated from tissues as described above. Unlike bacteria, most tissue cells are not adapted to living in suspension and require a solid surface on which to grow and divide, which is now usually the surface of a plastic tissue-culture dish (Figure 4–32). Cells vary in their requirements, however, and some will not grow or differentiate unless the culture dish is coated with specific extracellular matrix components, such as collagen or laminin.

Cultures prepared directly from the tissues of an organism, either with or without an initial cell-fractionation step, are called **primary cultures.** In most cases cells in primary cultures can be removed from the culture dish and used to form a large number of **secondary cultures;** they may be repeatedly subcultured in this way for weeks or months. Such cells often display many of the differentiated properties appropriate to their origin: fibroblasts continue to secrete

Figure 4–32 Cells in culture. Scanning electron micrograph of rat fibroblasts growing on the plastic surface of a tissue-culture dish. (Courtesy of Guenter Albrecht-Buehler.)